EDITORIAL

Anti-Covid 19 RNA Vaccines

By the end of 2019, an epidemic was slowly beginning in China in Wuhan, the capital of Hubei province. This, like others before, was caused by a coronavirus, named since SARS-CoV-2. Since then, this modest epidemic has evolved into a pandemic over the weeks, first spreading to South East Asia and the entire Far East and then spreading inexorably from East to West, becoming a pandemic in a matter of months, affecting all countries and resulting in more than 1.5 million deaths, mainly among the elderly and people suffering from various comorbidities, such as diabetes, overweight, high blood pressure, etc. In the absence of an effective treatment and awaiting for an hypothetical vaccine, the vast majority of countries choose to fight it by conventional weapons that the history of the numerous epidemics that punctuated that of humanity, taught us namely quarantine, isolation of affected people, decrease of personal contact and the wearing of masks, in short a whole series of gestures barriers and behavior aimed at limiting the spread of the virus which was all the more insidious with a large proportion , more than half, of those infected and contagious had no symptoms and in the absence of a diagnostic test were unaware of their contagiousness. These necessary precautions, in the absence of adequate treatment lead to the closure of schools, of stores deemed to be non-essential, a sudden halt in economic activity compensated for a small part by remote work and a significant increase in precariousness.

A search for treatment from molecules known for other uses has resulted in numerous or even too many clinical trials that unfortunately have failed to find the miracle cure outside of dexamethasone which significantly decreases the severity of respiratory symptoms. In parallel to these clinical trials the pharmaceutical industry and many biotechnology companies sought to develop a vaccine.

Vaccination is based on a general principle of presenting the body with proteins or protein fragments from the microorganism that we want to protect against by eliciting antibodies and cellular immunogenicity from the recipient to be ready to fight a subsequent infection.

To achieve this goal, many technological approaches have been developed, from the simplest of injecting the microorganism itself, which a pre-heat treatment or a chemical will have devoid of any pathogenicity, to the most recent one of injecting a fragment of RNA encoding a protein necessary for the development of the microorganism.

Thus, on November 9, 2020, Pfizer announced that it had developed a particularly effective RNA vaccine with the German company BioNtech, since intermediate Phase III analyses showed efficacy of more than 90%, a figure calculated by comparing the number of people who contracted the virus in the placebo group compared to the vaccinated group after two injections at three weeks intervals and this without significant side effects. Two months later, the hopes were confirmed and a provisional marketing authorization was granted by the health regulators of the United States, Great Britain and then Europe so that the first vaccinations

started everywhere, although at very different rates.
Since Pfizer's announcement, Moderna, a biotechnology company based in the United States, has also announced the arrival of a vaccine also based on messenger RNA and showing equivalent performance. In addition, Russian and Chinese vaccines having not demonstrated their efficacy and safety through conventional Phase I, II, III studies are nevertheless utilized but in geographical areas that does not concern us.

In France, any vaccination, whatever the objective, raises a greater proportion of mistrust than in any other countries for several years. For example, a survey conducted the day after Pfizer-BioNtech's announcement, 47% of our citizens thought they would not get vaccinated, whereas this is our only hope of escaping a more or less severe containment.
The usual criticisms and fears against the Pfizer-BioNtech vaccine from anti-vaccines and skeptics are compounded by a particular fear due to the technology used which was judged, particular and too new, as well as the speed with which this vaccine was developed.

Messenger RNA, a new technology. Is that correct?
Listening to the countless comments of the written and spoken press, the public gets the impression that this technology for the development of an RNA vaccine like those of Pfizer-BioNtech and Moderna has been imagined and developed since the emergence of Covid 19. This is entirely false. The interest in RNA as a drug source has been recognized for many years and the number of publications concerned with this subject is counting in the thousands with an acceleration over the last ten years. Finally, over the past 5 years, several scientific papers devoted to the development of RNA-based vaccines have been published [1-3]. If this sort of achievements has been delayed this is mainly due to the fragility of the RNA that when introduces into a cell is very quickly destroyed as well as due to the natural immunogenicity of RNA. It was therefore necessary to find technical biases to correct these defects. In the case of Pfizer BioNtech [4], the viral RNA fragment is synthesized in vitro in the presence of 1 methyl pseudouridine in place of uridine to decrease its immunogenic character and increase the efficiency of the cellular machinery for synthesizing the virus spike protein [5] that binds to the ACE2 cell receptor. In addition, the vaccine preparation provides the terminal domain of the T4 phage fibritin to obtain a better spatial configuration of the newly synthesized antigen and allowing the synthesis of antibodies with greater affinity [6]. Finally, to allow the RNA vaccine once injected not to be degraded before reaching its target, it is coated in lipid nanoparticles [7]. Thus far from being a novelty likely to make suspicious, reading the numerous publications on this subject and other related must reassure on this technology which also offers many advantages over its competitors.
In fact, as the injected RNA was shown to be able to direct the synthesis of the encoded protein, the only remaining question was whether this protein would be sufficiently antigenic to induce the synthesis of antibodies in quantity and whether they would be protective. However, this question is more a matter of the choice made of the antigen than of the mode of delivery. A question potentially in favor of past vaccine manufacturing techniques that using the entirety of a killed virus offer a wider range of potentially antigenic proteins that may elicit a greater variety of antibodies. Fortunately, the results of

Phase III produced by Pfizer BioNtech show that the choice of this antigenic target was wise. However, the question of the persistence of protection remains, as we have only a three-month decline to date.

On the other hand, we can only welcome the speed with which this vaccine was produced. There are a number of ways to explain this. The need and urgency to have a vaccine, the only weapon to combat this pandemic in the absence of a dedicated treatment, has resulted in an unprecedented mobilization of many competent pharmaceutical companies, with significant resources and well-established logistics as well as a large number of biotech companies of various sizes, agile and reactive. This has resulted in dozens of projects based on virtually all possible vaccine development techniques. Added to this competition was a significant financial risk-taking on the part of these companies but also of the states that subsidized part of this research and pre-ordered hundreds of millions of doses of vaccines that did not yet exist to several companies in parallel, in order to minimize the risk of failure and not to be deprived in case of bad choices. Another important point explaining the speed of the first vaccines being brought to market is the risk taken by the various companies not to wait until the end of each step to initiate the next but to overlap them at the risk of having to interrupt everything if the results of a previous step were not conclusive and also to start manufacturing the candidate vaccine before the authorization to market and even the end of Phase III. Finally, very early on, the health authorities were informed of the developments so that they were able to judge the quality and solidity of the results almost in real time. They were themselves very responsive in a way that pharmaceutical files that very frequently require years of back and forth between the company and the regulatory body were this time processed in a few weeks. However, it should be stressed that this speed should not be equated with any laxity or risk-taking on the quality of the vaccine. The risk-taking in this case was only financial that the urgency required to take. On the other hand, skipping or decreasing to the extreme a phase III as has been done in some countries is not permissible and cannot be justified by urgency.

Can an RNA vaccine alter the host genome?

The RNA nature of this vaccine has made to fantasize about the risk of a change in the host genome. Fear again unfounded. Indeed, once the viral RNA has penetrated the cells, it loses its lipid protection to be translated which, also gives it a high sensitivity to nucleases. Moreover, to integrate into the host genome it would have to be retrotranscribed to DNA by the action of a retrotranscriptase. Remember that if the HIV virus can integrate into the host genome, it is because it has a gene encoding its own retrotranscriptase, a gene that SARS-Cov2 lacks. In addition, this vaccine is made with only a small part of the virus genome.

The new mutants

To date, several hundred thousand mutations have been identified when comparing the new sequences to that obtained at the very beginning of the epidemic. To this, no wonder. Like all RNA viruses, SARS-Cov2 mutates a lot. Every time a viral particle replicates its genome, the reliability of the process is not absolute and errors occur This phenomenon is general, DNA, itself the custodian of our identity, is no exception, but our cells are equipped with several enzymatic systems that track integration errors and correct them, for the most part,

although not all and this is why the comparison of two human genomes reveals point differences in the order of one different nucleotide per 1000. On the other hand, RNA viruses lack a backup system and therefore accumulate mutations with each replication of their genome. The fate of these mutants then depends on the selective advantage or not that a mutation confers on this new variant. This depends on the mutation citself and the function of the mutated protein. If this mutation promotes the development of the virus and cause the carriers of this mutation to produce more particles, this virus will become more contagious which, will result in an increase in the number of people infected with this virus at the population level. It may also happen that the spike protein of the new mutant has a better affinity for the ACE2 cellular receptor. All this may explain the current dramatic development of the English mutant. Moreover, these mutations could alter the affinity of antibodies resulting from vaccination with a virus of the first wave. This does not seem to be the case at the moment. Although the two mutations called N501Y and P681H, because of their strategic location on the spicule protein and the chemical nature of the changes, a tyrosine instead of an asparagine in position 501 or a proline replaced by a histidine in 601, which from a structural point of view are not neutral raise concern. In addition, this new variant has a deletion of two previously observed amino acids 669-670 del that was beyond the immune system in an immunosuppressed patient.

All this to say that more efforts must be made to sequence more viruses and better understand the appearance and development of variants and that the longer the epidemic continues, the more we will be confronted with variants, which through the natural, universal game of selection will make us expect to have to deal with increasingly contagious viruses., but not necessarily more pathogenic or at least deadly, because it is not in the interest of a virus to kill its host before it develops.

In this context, RNA vaccines are likely to have advantages over traditional vaccines. Indeed, their method of preparation once controlled, it should be more easily adaptable to new variants by changing in the vaccine preparation the RNA used as a matrix to produce the desired antigen. This may to some extent explain the rapid success in the development and production of these RNA vaccines, the only ones authorized to date. So, if the know-how and experience in this approach are present in the companies, what was the case at NBiotech and Moderna, the roadmap is relatively simple and the development predictable, except to have made a bad choice regarding the antigenic protein, which is not currently the case. Analyses of the Phase III data also show the notable absence of significant side effects apart from some transient disturbances. Only people with allergies and especially those who have already had anaphylactic shock accidents should not receive these vaccines. For the rest, the decline is too short, except to point out that to date several million people have received the Pfizer BioNtech vaccine without a proportional increase in the number, variety or severity of side effects.

Finally, it is important to note that the Pfizer BioNtech's vaccine preparation does not include an adjuvant, which should silence some of the recurring criticisms of this issue.

Much has been said and written about the logistical difficulties of retaining these vaccines at -70 degrees. Of course, this is an additional problem in the delivery of

doses to vaccination sites. However, it is not reasonable to exaggerate it. Indeed, all biology laboratories, all researchers in the field regularly send and receive packages with products requiring this method of preservation. The problem is easily addressed by the addition of dry ice which allows for problem-free storage for several days in a klegecell box containing a few kg of dry ice, so the risk of vaccine loss for poor preservation should be zero.

Other questions arise for which it is too early to answer, but let us not spoil our pleasure at this long-awaited success. Finally, because of these successes, it is likely that RNA vaccines are promised a great future, especially since we are not immune to other pandemics.

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Francis Galibert, Emeritus Professor
Académie nationale de médecine, 16 rue Bonaparte, 75006 Paris, France
galibert@univ-rennes1.fr